



## Larval morphology and identification of *Rhyacophila meyeri* McLachlan 1879 (Trichoptera: Rhyacophilidae)

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### Abstract

This paper describes the previously unknown larva of *Rhyacophila meyeri* McLachlan 1879. Information on the morphology of the 5th larval instar is given, and the most important diagnostic features are illustrated. In the context of existing identification keys the larva of *R. meyeri* keys together with *R. stigmatica* (Kolenati 1859). The species pair can be separated by differences in the setation pattern at the anterior pronotal border and maximum head width.

With respect to zoogeography, *Rhyacophila meyeri* is reported from Switzerland and northern Italy whereas *R. stigmatica* is restricted to the Austrian, German and Swiss Alps and northern Slovenia (Cianficconi 2002; Graf *et al.* 2008; Lubini-Ferlin & Vicentini 2005; Malicky 2009; Robert 2004).

**Key words:** 5th instar larvae, caddisflies, morphology, key, distribution

### Introduction

In 1950, Döhler defined six groups of Central European *Rhyacophila* species (*Hyperhyacophila*, *Pararhyacophila*, *Hyporhyacophila*, *Metarhyacophila*, *Prosrhyacophila* and *Rhyacophila* s.str.), based on the number and shape of larval gills. Whilst this morphological concept is not substantiated by adult morphology (e.g., Schmid 1970), Döhler's classification nevertheless is commonly used to characterize larval morphology as this striking feature is one of the most reliable characters in distinguishing different taxa.

From the 35 *Rhyacophila* species so far reported from the Alps (ecoregion 4 *sensu* Illies 1978; Graf *et al.* 2008; Malicky 2009), the larvae of 8 species conform to Döhler's group *Hyporhyacophila* in which gills are completely lacking; *Rhyacophila glareosa* McLachlan 1867, *R. hirticornis* McLachlan 1869, *R. philopotamoides* McLachlan 1879, *R. stigmatica* (Kolenati 1859), *R. aquitanica* McLachlan 1879, *R. pubescens* Pictet 1834, *R. tristis* Pictet 1834 and the hitherto unknown larva of *R. meyeri* McLachlan 1879. Larval and adult material of the latter species was recently collected in Valchiusella (Italy). Whether the unknown larvae of four further species (*R. schmidinarica* Urbanič *et al.* 2000, *R. orobica* Moretti 1991 [both *R. philopotamoides* Group; Botosaneanu 1999; Moretti 1991; Urbanič *et al.* 2000], *R. kelnrae* Schmid 1971 and *R. konradthaleri* Malicky 2009 [both *R. stigmatica* Group; Malicky 2009; Schmid 1971]) conform to Döhler's group *Hyporhyacophila* remains unknown due to a lack of material.

Descriptions of hitherto unknown larvae are especially important because furthering our species-level taxonomic knowledge will directly lead to an improved ecological understanding of their habitats which, in turn, is a prerequisite for defining basic environmental data for stream assessment procedures. Contemporary biodiversity research crucially depends on the definition and identification of species as evolutionary units within classification systems. Shortcomings in identification may not only underestimate faunal diversity but bias ecological assessment procedures and may question the comparability of data within intercalibration processes as well as the validity of assessment systems *in toto*.

## Material and methods

Six adults (5 males, 1 female), 4 final instar larvae, and 1 prepupa of *Rhyacophila meyeri* were collected by W.G. and S.V. on 21 June 2014 using a hand net and a light trap at a spring brook in Valchiusella near Traversella, Tisone (Italy), at 1200 m above sea level (45° 32' 11.8" N, 07° 40' 47.8" E) and preserved in 70% ethanol. The larvae were studied and photographed using a Nikon SMZ 1500 binocular microscope with DS-Fi1 camera and NIS-elements D 3.1 image stacking software for combining 8–45 frames in one focused image.

Deposition of voucher specimens: Two 5th instar larvae of *Rhyacophila meyeri* are deposited in the collection of J. Waringer (Vienna, Austria), two 5th instar larvae, one prepupa, five males and one female in the collection of W. Graf (Vienna, Austria). In addition, the following comparative material has been used (collection of J. Waringer, Vienna, Austria): *R. glareosa* (five 5th instar larvae), *R. hirticornis* (seven 5th instar larvae), *R. philopotamoides* (one 5th instar larva), *R. stigmatica* (three 5th instar larvae), *R. tristis* (seven 5th instar larvae) and *R. pubescens* (three 5th instar larvae).

Species affiliation was enabled by the fact that putative *Rhyacophila meyeri* larvae were collected within the species-specific geographical range of this taxon, based on the revision of the alpine species of the *R. stigmatica* Group by Malicky (2009), and by the fact that adults of both sexes were collected at the same sites as the unknown larvae.

## Results

### Description of the fifth instar larva of *Rhyacophila meyeri*

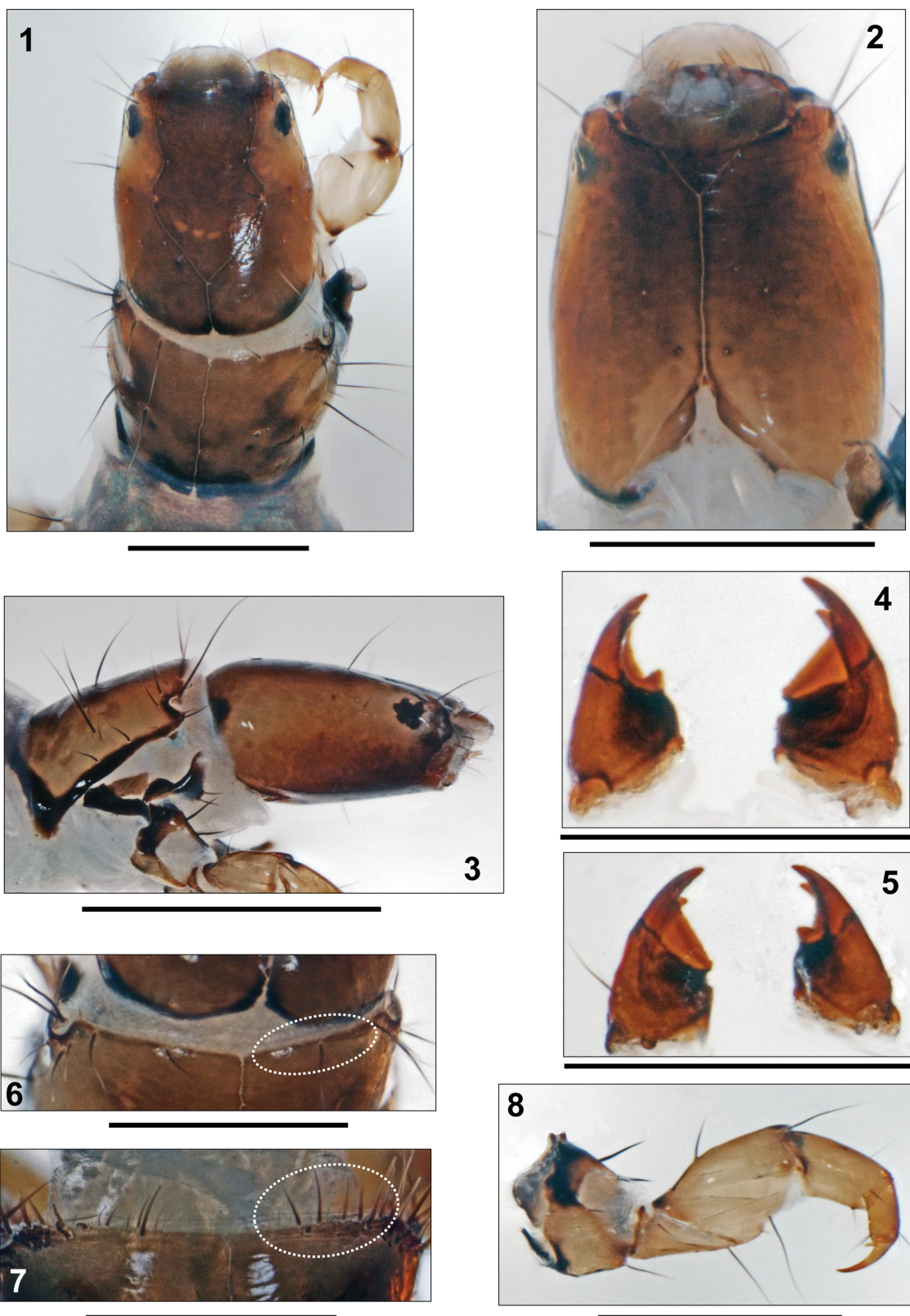
Morphological terminology after Wiggins (1998).

**Biometry.** Body length of final instar larva ranging from 9.5 to 11.2 mm, head width from 0.55 to 0.68 mm (n=4).

**Head.** Head capsule prognathous, elongated, with smooth surface and maximum width at head center (Fig. 1). Dorsal and ventral head sections chestnut brown, with pale muscle attachment spots on frontoclypeus and brown ones on parietals (Figs. 1–2). With paler areas around eyes, along lateral sections of parietals and ventral of occipital margin (Figs. 1–3). Head capsule with complete set of primary setae (5 pairs on frontoclypeus, 11 pairs on parietals), with setae # 6, 7, 9 and 14 long and conspicuous. In addition, 13 pairs of sensory pits (sensilla campaniformia) are present, 4 pairs of them situated on frontoclypeus. Frontoclypeus bell-shaped, with deep central constriction posterior of eye level and with bulge within posterior third (Fig. 1). Antennae near anterior parietal border, extremely small. Sclerotized labrum semicircular, yellowish brown with pale anterior border (Fig. 1). Submentum dark brown, triangular, distinctly wider than long (Fig. 2). Mandibles medium brown, almost black centrally, and asymmetrical (Figs. 4, 5): in dorsal view, left mandible with short, stout basal tooth (lacking on right mandible and replaced there by straight cutting edge). Both mandibles with apical and subapical teeth, distinctly larger on right mandible. Hairy brush in central concavity lacking. Each mandible base with one very short and one large dark seta (Fig. 5).

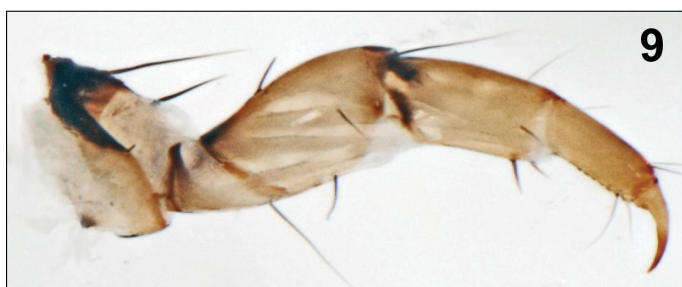
**Thorax.** Pronotum fully covered by 2 large sclerites, tapering posteriorly, wider than head capsule; pronotal sclerites chestnut brown, slightly lighter in colour laterally, with black or brown muscle attachment spots creating arc-like patterns, their arms (1) meeting centrally, (2) pointing towards anterolateral corners and (3) pointing towards posterior border (Figs. 1, 3). Posterior pronotal corners slightly prolonged ventrally. Posterior and lateral margins thickened and darkly striped, including anterolateral corners. Pronotal notch at anterolateral corner large, deep and semicircular, with one large and two smaller setae. Up to 10 setae in each anterolateral group surrounding pronotal notch. Anterior borders of pronotal sclerites with dense fringe of very short, curved, fine, yellow setae and 2 long primary setae (Fig. 6, dotted oval). In addition, 7 setae of varying lengths distributed over central section of each pronotal half. Prosternal horn lacking.

Mesothorax dark purplish-brown to purplish-blue with coloured areas subdivided by whitish bands (Fig. 11). Mesonotum largely unsclerotized except small roundish sclerotized plate at dorsal posterior center (Fig. 11, dotted oval). Metathorax same colour, completely unsclerotized. Foretrochantin large, with dark central stripe and large rounded anterior process (Fig. 3). Mid- and hind trochantin smaller, elongated, with blackish ventral margin.



**FIGURES 1–6.** *Rhyacophila meyeri* McLachlan 1879, 5th instar larva. **1**, Head and pronotum, dorsal. **2**, Head, ventral. **3**, Head and pronotum, right lateral. **4**, Mandibles, dorsal. **5**, Mandibles, ventral. **6**, Detail of anterior pronotal border, dorsal (dotted oval: only short, curved setae and few long primary setae present). **FIGURE 7.** *Rhyacophila stigmatica* (Kolenati 1859), 5th instar larva. Detail of anterior pronotal border, dorsal (dotted oval: short, curved setae, widely-spaced intermediate and few long primary setae present). **FIGURE 8.** *Rhyacophila meyeri* McLachlan 1879, 5th instar larva. Right foreleg, posterior face. Scale bars: 0.5 mm (except 3: 1 mm).





**FIGURES 9–12.** *Rhyacophila meyeri* McLachlan 1879, 5th instar larva. **9**, Right midleg, posterior face. **10**, Right hind leg, posterior face. **11**, Meso-, metanotum and anterior abdominal segments, dorsal (dotted oval: sclerite at mesonotum). **12**, Tip of abdomen, right lateral (s= dorsal sclerite on abdominal segment IX; arrow: teeth on anal claw). **13**, Sampling site (spring brook in Valchiusella near Traversella, Tisone, Italy; 1200 m above sea level). Scale bars: 0.5 mm (except 11: 1 mm).

Legs pale brown, with numerous setae on coxae, trochanters and femora; tibiae and tarsi with only small number of setae (Figs. 8–10). Forefemora distinctly wider than mid- and hind femora. Each tibia with 2 long, brown, slender apical spurs. Tarsal claws long and slender, curved, pointed, with basal spur (Figs. 8–10).



**Abdomen.** Abdomen slightly flattened dorsoventrally; dorsally dark purplish-brown to purplish-blue, with coloured areas subdivided by whitish bands. Laterally and ventrally creamish white (Fig. 11). Gills and lateral fringe lacking.

Abdominal dorsum IX with large, yellow quadrangular sclerite with medium brown band along anterior border (Fig. 12s). Anal proleg lateral sclerite without curved spike, but with freely projecting ventral hook. Anal claw long, regularly curved, partially divided by ventral membranous section and fitted with 2–3 ventral teeth (Fig. 12, arrow).

### **Morphological separation of fifth instar larvae of *Rhyacophila meyeri* McLachlan 1879 from other European Trichoptera**

A summary of morphological features for the identification of Rhyacophilidae larvae was provided by Buholzer (1978), Coppa *et al.* (2012), Edington & Hildrew (1995), Graf (1993), Viera-Lanero (2000), and Waringer & Graf (2011, 2013). Within the framework of the above Rhyacophilidae keys, the genus *Rhyacophila* is separable from other caddis larvae by the following combination of characters:

- pronotum completely sclerotized, meso- and metanotum unsclerotized except a tiny dorsal sclerite which is present in some *Rhyacophila* species at the center of the posterior mesonotal border (Figs. 1, 11);
- without transportable case;
- prosternal horn lacking;
- labrum sclerotized (Figs. 1, 2);
- abdominal segment IX with dorsal sclerite (Fig. 12s);

Within the genus *Rhyacophila*, gill filaments are completely lacking in larvae of *R. meyeri* (Fig. 11) as in other species of the *Hyporhyacophila* group for which larvae have been described (*Rhyacophila glareosa*, *R. hirticornis*, *R. philopotamoides*, *R. stigmatica*, *R. aquitanica*, *R. pubescens*, *R. tristis*). Based on the presence of ventral teeth at the anal claw, *R. meyeri* keys together with *R. glareosa*, *R. hirticornis*, *R. philopotamoides*, and *R. stigmatica*. Within this group, the ventral side of the head capsule is pale in *R. glareosa*, *R. hirticornis*, and *R. philopotamoides* whereas it is dark in *R. meyeri* and *R. stigmatica*. The latter species pair can be separated morphologically by the fact that in *R. meyeri*, intermediate dark setae are lacking at the anterior pronotal border, and biometrically by the maximum head width of the last instar larvae which is 0.55–0.68 mm in *R. meyeri* but  $\geq 0.90$  mm in *R. stigmatica* (Table 1).

### **Discussion**

McLachlan (1879) described *Rhyacophila meyeri* based on Swiss material collected by Meyer-Dür at Gadmen (1400 m above sea level). Although the taxonomic status and geographic range of *R. meyeri*, a member of the *R. stigmatica* Group, was thought to be well established, Malicky (2009) was able to show that part of the specimens attributed to *R. meyeri* differed in the structures of the distal section of abdominal segment X, the phallosome, the aedeagus, the fingerlike processes of segment IX and the second segment of the lower appendages and that those specimens were in fact a new species (*R. konradthaleri*). According to these findings, *R. meyeri* is now restricted to Switzerland and Italy, endemic to an area stretching approximately between western Lombardia, the Gadmen valley, and the Sarntal Alps ('insubric endemite' *sensu* Malicky 2009) whereas *R. konradthaleri* is known from Austria (Carinthia, Styria, Upper Austria) and northern Slovenia in a stretch from the Pyhrn pass in the north to the Pohorje mountains in the south. Together with the range of *R. keltnerae* (France: Val Boréon; Liguria, Piemonte in the north to the Alpi Apuane in the southeast), the geographical ranges of those three *Rhyacophila* species are hypothesized to be identical with some of the most important refugia during the Pleistocene (Malicky 2009).

The larvae, prepupa and adults of *Rhyacophila meyeri* were sampled on 21 June 2014 by Wolfram Graf and Simon Vitecek; adults are known to be on the wing until 13 November (Italy: Piemonte; Zobodat 2015). The species inhabits oxygen-rich montane and subalpine habitats like springs and high-gradient springbrooks fitted with hard substrate sediments (coarse gravel, cobbles, boulders and bedrock; Graf *et al.* 2008).

**TABLE 1.** Synopsis of characters separating the currently known larvae of family Rhyacophilidae (5th instars) without gills (group *Hyporhyacophila sensu* Döhler 1950) from the Alps (ecoregion 4; Graf *et al.* 2008).

Species/character	Anal claw with ventral teeth	Colour of ventral head surface	Maximum head width	Head width (mm)	Colour of muscle attachment spots at dorsal head surface	References
<i>Rhyacophila glareosa</i>	yes	light	head center	0.90	without spots	Buholzer 1978
<i>Rhyacophila hirticornis</i> <sup>1</sup>	yes	light	head center	1.20–1.30	dark	Buholzer 1978
<i>Rhyacophila philopotamoides</i> <sup>1</sup>	yes	light	head center	1.00–1.10	dark	Buholzer 1978
<i>Rhyacophila meyeri</i> <sup>2</sup>	yes	dark	head center	0.55–0.68	pale	Present paper
<i>Rhyacophila stigmatica</i> <sup>2</sup>	yes	dark	head center	0.90–1.08	pale	Graf 1993
<i>Rhyacophila aquitana</i>	no	dark	head center	1.10–1.20	pale	Coppa <i>et al.</i> 2012
<i>Rhyacophila pubescens</i>	no	light	head parallel-sided	1.00–1.10	pale	Coppa <i>et al.</i> 2012
<i>Rhyacophila tristis</i>	no	dark	posterior third	1.10–1.20	pale	Coppa <i>et al.</i> 2012

<sup>1</sup> In *R. hirticornis*, a small sclerotized patch is present at the dorsal posterior center of the mesothorax (Fig. 11, dotted oval) which is lacking in *R. philopotamoides*.

<sup>2</sup> In *R. stigmatica*, setation at the anterior pronotal border consists of (1) a dense fringe of very short, curved, fine, yellow setae, (2) a widely-spaced row of intermediate dark setae and (3) few long primary setae (Fig. 7); in *R. meyeri*, intermediate dark setae are lacking (Fig. 6).

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